Mode of Action of Plastic Film in Extending Life of Lemon and Bell Pepper Fruits by Alleviation of Water Stress¹

Received for publication December 22, 1982 and in revised form April 18, 1983

SHIMSHON BEN-YEHOSHUA, BORIS SHAPIRO, ZEV EVEN CHEN, AND SUSAN LURIE

Department of Fruit and Vegetable Storage, Agricultural Research Organization, Bet Dagan 50250, Israel

ABSTRACT

The mechanism by which seal-packaging individual fruit in high density polyethylene film delays deterioration was investigated with lemon (Citrus limon [L.] Burm. f. cv Eureka) and bell pepper (Capsicum annuum L. cv Maor) fruits. Seal-packaging effects were due to the watersaturated atmosphere in the sealed enclosure around the fruit. Softening of fruit was highly correlated with declining water potential of fruit. Sealing drastically inhibited softening as well as changes in cell wall pectins. Sealing also delayed disintegration of membrane as shown by the inhibited leakage of amino acids, in particular, and electrolytes in general. All these effects of sealing were prevented or reduced by including hygroscopic CaCl2 in the sealed enclosure which reduced the ambient humidity. Furthermore, some of these effects of sealing could be achieved also by maintaining nonsealed fruit in water-saturated atmosphere. Sealing effects could not be related to a possible 'modified atmosphere' mechanism in O2, CO2, or ethylene. This work supports the hypothesis that the mode of action of sealing in the polyethylene relates to the alleviation of water stress which exists in harvested fruit.

Among the various techniques developed to extend fruit postharvest life, the use of plastic film is growing in importance because it is convenient in the many different conditions throughout the chain of handling from producer to consumer (8, 21). Ben-Yehoshua (8) found that seal-packaging individual fruit with a thin film of HDPE², 5 to 15 μ m in thickness, doubled storage life of various fruits. The flavor of the fruit was not spoiled; on the contrary, the sealed fruit maintained its normal flavor, as well as its firmness and fresh appearance for at least twice as long as nonsealed fruit. Surprisingly, sealed citrus fruit kept at 20°C lost less weight and was firmer than nonsealed fruit at optimal (lower) temperatures.

Seal-packaging delayed several parameters of physiological deterioration of fruit better than cooling; which is usually considered the best means of delaying senescence of detached living organs (11, 21). Accordingly, it is hoped that the elucidation of the mode of this action of seal-packaging would also help better understand the mechanism of senescence of detached plant organs and their causes. Such information might also be useful in extending the basic knowledge of the control of the deterio-

ration of other perishable, living fruit items.

The control of deterioration of fruit by plastic film had been generally explained by the modification of the concentrations of CO_2 , O_2 , and ethylene (21). However, this theory was not supported by our data (10, 13). Endogenous levels of the above gases were similar in fruit sealed in HDPE or not sealed, but their rates of physiological deterioration were altogether different. The major effect of sealing appears to be the provision of a watersaturated microatmosphere (8). This may alleviate water stress, which is important in controlling life processes (15, 20). However, so far only the inhibition of transpiration, shrinkage, and shriveling has been ascribed to the high humidity that is found in the microatmosphere of the sealed fruit (21). Therefore, we set out to investigate the hypothesis that sealing delays deterioration of lemon and bell pepper fruits by alleviating water stress. The importance of transpiration as a major process leading to physiological deterioration of citrus fruit has been demonstrated (7). This paper reports our attempts to relate water stress to various physiological parameters in sealed and nonsealed fruit. Lemons and peppers were selected for these studies because they responded very favorably to the seal-packaging technique. These fruits do not have the marked ripening and climacteric processes that could disturb our objective by masking the effects of the sealing per se on fruit behavior.

MATERIALS AND METHODS

Lemon (Citrus limon [L.] Burm. f. cv Eureka) and bell pepper (Capsicum annuum L. cv Maor) fruits were obtained directly from orchard or field, or from packing houses before storage. Samples of fruit of uniform size and appearance, originating in one orchard, were subjected to different treatments. Normal treatment for both lemon and pepper was storage for various lengths of time at 85% RH and steady temperature, 14 or 20°C for lemon and 17 or 8°C for pepper, either nonsealed or sealed in HDPE (Mitsui, Tokyo, Japan). The film was applied and sealed around the fruit with a unit manufactured by Swery Electronics Inc. Israel.

In one pepper experiment, two other treatments were performed: WSA and sealed atmosphere with around 85% RH. The WSA was obtained by placing nonsealed peppers in a Plexiglas chamber with a humidity-generating machine at the bottom. The amount of humidity was regulated by Lambrecht humidity regulator, and was independently checked periodically by a calibrated thermohydrograph. The humidity inside the sealed enclosure was measured by a Duratherm hygrometer (Environmental Tectonics Corp., Southampton, PA) that has accuracy of ±2% RH. The sealed atmosphere that was supposed to have 85% RH was arranged by introducing 5 g of CaCl₂ drying crystals into the sealed HDPE bag with the pepper. The CaCl₂ was changed weekly. The humidity in this sealed bag increased from 80 to 88% over the week, while the HDPE-sealed pepper without CaCl₂ registered a steady 97% RH (as measured with a hair hygrometer).

¹ Supported by a grant from the United States-Israel Binational Agricultural Research Development Fund. Contribution from the Agricultural Research Organization, The Volcani Center, Bet Dagan, Israel. No. 517-B, 1982 series.

² Abbreviations: HDPE, high density polyethylene; WSA, water-saturated atmosphere; WSD, water saturation deficit; AIS, alcohol-insoluble solids.

Table I. Effect of Seal-Packaging in HDPE on Weight Loss, Firmness, and Water potential of Lemons Kept at 17°C and 85% RH

Time after Harvest	HDPE Sealed	Weight Loss	Firmness (Deformation)	Water Potential
months		% of initial	mm	bars
0		0.0	3.0	-13.7
1	_	5.7	5.2	-15.2
	+	0.8	3.1	-13.4
2	_	10.3	7.0	-21.1
	+	1.2	3.5	-15.0
b nonsealed ^a		5.15*	2.0*	−3.70 *
b sealed Significance of difference between		0.60 ns	0.25 ns	-0.65 NS
the two coefficients		*	*	*

^{*} Regression analysis was carried out of the nonsealed and sealed treatments over time and the significance of each of the regression coefficients (b) as well as the differences between the two regression coefficients (b) was tested at 5% level. * is significant.

The following parameters were studied on all types of fruit: weight loss; firmness; water deficit and tissue water potential; membrane integrity as measured by electrolyte and amino acid leakage; cell wall properties; CO₂ and ethylene production; and endogenous content of O₂, CO₂, and ethylene. Samples were of the size required for statistical analysis, *i.e.* five to 10 replicates for each measurement.

Weight Loss and Firmness. Ten fruit from each treatment were weighed at the beginning of the experiment and at various times during the course of storage, and the results were expressed as percentage of loss from initial weight. Fruit firmness was determined with a compression tester (7, 8), using a 5 kg weight for lemons and 2 kg for peppers. Full deformation was measured 30 s after exerting the force on the fruit, then the weight was removed and residual deformation was measured 15 s later. The firmer the fruit, the lower were both readings. The correlation between these two measurements for bell pepper was 0.99.

Water Deficit and Tissue Water Potential. The status of water in the fruit was measured in two independent ways (3): (a) water potential expressing the free energy of the water molecule and considered the final determinant of diffusional water movement; and (b) WSD that expresses the relative water content of the tissue and measured according to the formula:

$$WSD = \frac{Saturated weight- original fresh weight}{Saturated weight- oven dry weight} \times 100$$

The water potential was measured on discs from fruit using a J14 leaf press (Campbell Scientific Inc, Logan, UT). This new instrument for studying the water potential has many advantages over the Scholander pressure chamber which cannot altogether be utilized for harvested fruit along its storage. Discs of the fruit were pressed and the reading was made at the appearance of the first sign of sap at the cut edges (16). Five fruit were measured for each treatment; three discs from each fruit. Results for discs from the same fruit were generally very similar. The leaf press was calibrated each time by randomly selecting eight of the fruit to be sampled and measuring them with both the leaf press and the gravimetric method. In the latter technique tissue discs are weighed, allowed to equilibrate in a range of sucrose solutions at room temperature for 1 h, and then reweighed. The tissue water potential is equal to the water potential of a sucrose solution in which the weight of tissue segments did not change. These values were compared with the leaf press measurements obtained on discs from the same fruit and a factor calculated to convert the

leaf press measurements of the fruit from p.s.i. to bars.

Membrane Integrity. Discs from pericarp of peppers or peel of lemons were put into 10 ml of distilled H₂O and incubated for 24 h on a shaker at 4°C. The discs were then ground in another 10 ml of distilled H₂O and filtered through glass wool. The incubation medium and the ground tissue were assayed for amino acids (32) and electrolytes using an El Hama TH 27 Conductometer (Israel). The results were expressed as percentage of total electrolytes or amino acids.

Cell Wall Contents. After the above parameters were measured, five samples from each treatment, 50 g each, were frozen for assay of cell wall. AIS were prepared from the frozen tissue (4). The resulting powder was dried and stored in HDPE-sealed bags until further analysis. Samples of 100 mg each were then extracted consecutively to obtain three fractions, water-soluble pectin, EDTA-soluble pectin, and insoluble pectin (4). The latter was digested with commercial pectinase (Sigma) to release the insoluble pectin fractions (27). The total amount of pectic sustances was determined following enzymic digestion on a separate 100-mg sample of the AIS preparation and was found to equal within 10% the sum of the above fractions. The quantity of the pectic material in each fraction was determined by the colorimetric carbazole method (30). The residue from the total pectin extraction was extracted first for 2 h at 100°C in 5% NaOH to release a hemicellulose fraction, and then the residue was treated according to Updegraff to obtain the cellulose fraction (33). Carbohydrates were determined by the phenol-sulfuric acid method (19).

Polygalacturonase and Cellulase Activity. About 50 g of fruit tissue were ground in 30 ml water containing 2.5 g NaCl for 2 min. The slurry was then filtered through cheesecloth and the resultant filtrate was used as enzyme crude extract. Enzyme activity was measured by viscosimetry using 1% citrus pectin for polygalacturonase activity and 1% carboxyl methyl cellulose for cellulase activity. The decrease in viscosity over 30 min was monitored.

Production of CO_2 and Ethylene and Their Endogenous Concentration. CO_2 and ethylene production were investigated with single fruit (7, 8). Inasmuch as C_2H_4 readings were at times too low, the C_2H_4 was absorbed with mercuric perchlorate and then released with HCl. Endogenous content of O_2 , CO_2 , and ethylene was measured (7, 8).

RESULTS

Effects on Firmness, Weight Loss, Water Potential, and Water Deficit. Sealing individual lemons in HDPE film delayed signif-

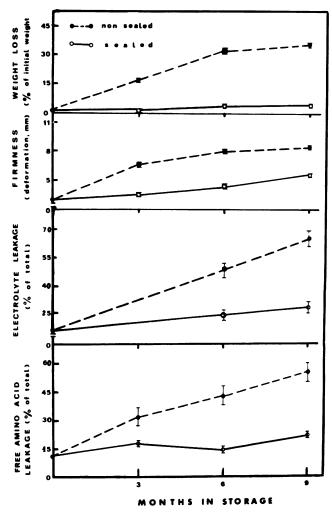


FIG. 1. Effect of seal-packaging in HDPE on lemons stored at 14°C and 85% RH for 9 months. (• – – •), Nonsealed; (O — O), sealed. SD is indicated.

icantly (at 1% level) weight loss and preserved firmness as expressed by deformation (Table I; Fig. 1). During storage non-sealed lemons lost 10 times more weight than HDPE-sealed ones. It was also found that the sealed fruit preserved its initial firmness for a long period of storage, while nonsealed lemons significantly softened. Similar changes were found when water potential in the peel was measured (Table I). Nonsealed lemons demonstrated lower values of water potential than HDPE-sealed fruit. In general, there was a strong correlation between the water status and the firmness of the fruit.

Similar results were obtained with bell peppers (Fig. 2). Non-sealed peppers lost 10 times more weight than sealed fruit during 4 weeks of storage at 17°C and 85% RH. The sealed peppers preserved their initial firmness while nonsealed ones softened and shrivelled. Two independent measurements of tissue water status, WSD and water potential, correlated highly with weight loss (r = 0.91 and 0.94, respectively). The values of WSD and water potential did not change from the time of harvest through the 4 weeks' storage in sealed fruit, while lower values were found in nonsealed fruit as a result of water stress. In some cases (data not shown), HDPE-sealed peppers retained their initial firmness even for 10 weeks of storage, and lemons stayed firm for a period of up to 1 year. The correlation coefficient between weight loss and firmness of pepper was 0.97 and between water potential and firmness, 0.98.

Effects on Membrane Integrity. Ripening or senescence of

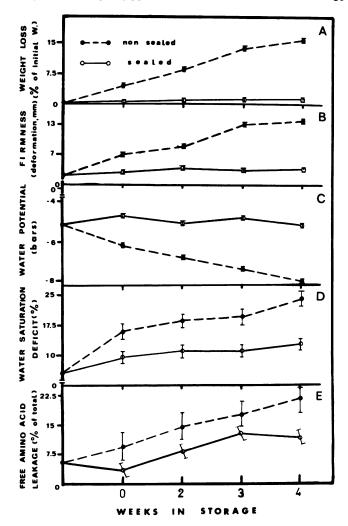


FIG. 2. Effect of seal-packaging in HDPE on green peppers stored at 17°C and 85% RH for 4 weeks. A, Weight loss; B, firmness; C, water potential; D, water saturation deficit; E, leakage of free amino acids. (O——O), Nonsealed. (O——O), sealed. SD is indicated.

detached plant organs is accompanied by the loss of membrane integrity (6, 31). One of the first cellular processes affected by water stress is membrane function (23). Sealed peppers and lemons demonstrated better membrane integrity as expressed by reduced values of free amino acid and electrolyte leakage (Figs. 1 and 2), The degree of leakage in nonsealed fruits at the end of the storage period was more than twice as high as that in sealed lemons and peppers.

Effect of WSA and Ambient RH. The hypothesis that sealpackaging extends storage life of fruit by preventing the formation of water stress that develops in nonsealed fruit during storage, was further tested by using WSA. Peppers were stored in a WSA and compared to HDPE sealing. As expected, weight loss was much inhibited in WSA and the decrease in WSD and water potential were prevented (Table II). Another treatment that could check whether the alleviation of water stress is the mode of action of seal-packaging was by studying whether producing water stress in sealed fruit would reduce its desirble action. By introducing hygroscopic CaCl₂ to HDPE-sealed peppers, RH inside the bags was reduced and stress could be produced. The reduction of the humidity inside the enclosure of the sealed fruit caused the expected changes; that is, the rate of weight loss and WSD was increased and water potential was lowered, similarly to changes occurring in nonsealed peppers. In addition, both firmness and membrane integrity were affected by the changes

Table II. Effect of RH and Seal-Packaging in High-Density Polyethylene Film on Firmness, Membrane Integrity, Water Potential, and Water Saturation Deficit of Green Bell Pepper Fruit Kept for 4 Weeks at 17°C

	Treatment				
Parameter Examined			Nonsealed	Sealed in HDPE + CaCl ₂ ^a	
Weight loss, %	1.76 ^b	1.2a	15.9d	10.5c	
Firmness, mm deformation	4.6b	3.3a	12.5d	9.9c	
Amino acid leakage, %	14.4ab	11.3a	21.5b	17.3ab	
Electrolyte leakage, %	21.6ab	19.2a	22.7b	22.3ab	
Water saturation deficit, %	11.5a	12.7a	24.4b	20.9b	
Water potential, bars	-5.2a	-5.2a	-8.0b	-7.5b	

^a Each fruit sealed in a plastic bag containing 5 g of CaCl₂ crystals.

Table III. Changes in Cell Wall Components of Green Pepper Fruit Stored at 17°C and 85% RH

		Galacturonic Acid or Glucose/AIS				
Time	HDPE Sealed	Water- soluble pectin	EDTA- soluble pectin	Insoluble pectin	Hemicellulose	Cellulose
weeks				μ/mg		
0		33.6	37.5	94.3	18.5	108.0
3	_	41.6	37.0	85.9	16.0	110.1
	+	37.0	40.2	88.4	17.0	101.4
4	_	50.2	44.0	81.4	14.0	118.3
	+	43.6	41.9	88.3	16.0	113.1
nonsealeda		3.81*	1.21 NS	-3.13*	-1.06 NS	2.14 NS
sealed		2.18 NS	1.05 NS	-1.61 NS	-0.60 ns	0.47 NS
Significance of difference between the two coefficients		*	NS	*	NS	NS

^a Regression analysis was carried out of the nonsealed and sealed treatments on time and the significance of each regression coefficient (b) as well as the differences between the two coefficients were tested. * is significant at 5% level.

of the RH. Firmness was maintained in the WSA and reduced by lowering the humidity around the seal-packed peppers. The leakage of both electrolytes and free amino acids was decreased in WSA and promoted by reduction of RH, although the differences were not always significant statistically. In general, treatments that preserved higher water potential in the pepper tissue, i.e. seal-packaging and WSA reduced membrane leakage. Whereas treatments which induced tissue water stress as evidenced by lower water potential, i.e. nonsealed and seal packaged with hygroscopic material, enhanced membrane leakage. The correlation coefficient between water potential and amino acid leakage was 0.91.

Inasmuch as the water content of the pepper fruits was closely related to fruit firmness, the possibility of restoring the firmness of soft fruit by placing it in aerated water was investigated. A group of 25 soft fruit that had a deformation of 9.0 mm were immersed in aerated water for 24 h. Their firmness rose to a deformation level of 6.0 mm which represented a recovery of 55% as compared to the initial firmness of 3.3 mm prior to softening. Fruit firmness could only partially be restored. Fruit already shrunken and wrinkled did not recover firmness.

Effect on Cell Wall Components. The cell wall components which showed changes during storage were mainly those of pectins (Table III). In the nonsealed pepper, the water-soluble pectin increased while the insoluble pectin decreased (significant at 5% level). In HDPE-sealed pepper, a smaller increase in these two fractions was found. The difference between sealed and non sealed ones was significant. There was no significant change in

the levels of cellulose and hemicellulose fraction during storage of the nonsealed fruit.

Parallel to the study of cell wall components, the activities of cellulase and polygalacturonase were examined in pepper fruit during storage (Table IV). No differences in cellulase activity was detected when sealed and nonsealed peppers were compared. This is in agreement with the finding that the levels of cellulose in the cell wall do not change during storage. On the other hand, sealing caused a decline in polygalacturonase activity during the course of storage, while the enzymic activity in nonsealed peppers remained unchanged. The results were that during storage the polygalacturonase activity was around 1.5 times higher in nonsealed peppers compared to sealed ones, which correlated well with the increasing levels of water-soluble pectin and EDTA-soluble pectin found in nonsealed peppers.

Effect on CO₂ and Ethylene Production and Their Endogenous Concentrations. The values of production and endogenous content were rather low and variable. However, the trend was that seal-packaging did not affect the CO₂ or ethylene production of lemons (Fig. 3) or their endogenous concentrations (Table V). Similarly, for green pepper kept at 17°C for 1 week, both respiration (7.5 ml CO₂ produced/kg·h) and ethylene production (0.04 μ l/kg·h) were similar for both sealed and nonsealed fruit. The endogenous concentrations of O₂ (20.5%), CO₂ (0.9%), and ethylene (0.03 μ l/l) were similar for both sealed and nonsealed fruit at 17°C with a tendency for the sealed fruit to have slightly, but insignificantly, higher CO₂ and C₂H₄ and lower O₂.

^b Mean separation by Duncan's multiple range test, 1% level.

HDPE Polygalac-Time Cellulase Sealed turonase weeks % activity/mg protein 1/30 min 0 139 20.0 1 17.4 21.0 17.5 16.8 2 156 21.6 15.6 15.9

15.4

14.6

11.6

11.4

-0.66 NS

-0.79 ns

NS

Table IV. Changes in Cellulase and Polygalacturonase Activities in Green Papper Fruits Stored at 17°C and 85% RH

Table V. Effects of HDPE Seal-Packaging on the Level of Ethylene and CO₂ Inside Lemons Kept for 4 Days at 20°C and 85% RH

3

Significance of difference between

the two coefficients

b nonsealed^b

b sealed

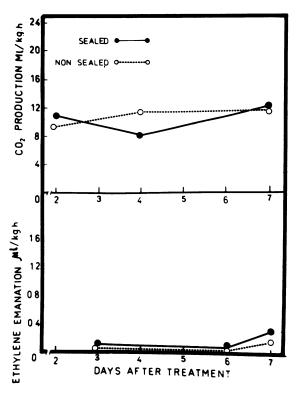
Sealing Treatment ^a	Ethylene	CO ₂	
	μl/l	%	
_	0.14a	0.53a	
+	0.14a	0.73a	

^a Five fruit per treatment; table represents a typical experiment out of five. Differences between the two packagings were found to be insignificant at 5% level with Student's *t* test.

DISCUSSION

The objective of this work is to relate the effects of sealing on lemons and bell peppers with those of water stress in these fruit and with the large body of data on water stress of detached organs (15, 20, 22, 24, 34, 35). Our results substantiate the hypothesis that the mode of action of the seal packaging in delaying senescence and maintaining firmness of these fruit is in the provision of WSA that alleviates water stress. The following observations support this theory. (a) The very high correlation between weight loss, firmness, water potential, and amino acid leakage, that is, effects of sealing on fruit deterioration and on water deficit are at least parallel. (b) Placing pepper in WSA delayed softening and the disintegration of membranes which water stress accelerated. Furthermore, firmness of bell pepper could be partly restored by immersion of a soft fruit in aerated water. (c) Reducing the RH of the microatmosphere of sealed fruit by the CaCl₂ cancelled the effects of sealing in delaying the disintegration of membranes and the softening process.

The observations conform with the many studies summarized by Boyer (15) that water stress affects tissues in the same manner as senescence. Leopold *et al.* (23) related water stress, membrane integrity, and solute leakage, and suggested also that leakage of desiccated leaves of cowpea reflects the condition of the cellular membrane. Similar findings were reported by others (26). It



20.2

12.9

17.2

12.4

-0.64 NS

-1.91*

Fig. 3. Effect of HDPE seal-packaging on ethylene and CO_2 production by lemon fruit. Fruit was waxed by Britex and kept at 20°C and 85% RH.

could be argued that leakage as well as other senescence phenomena occurred in parallel with the decline in water content, but were not caused by water stress. After all, this argument could continue, all these changes are the normal course of events in detached organs. However, the demonstrations that WSA per se

^a The polygalacturonase activity was based on 10 mg protein because of the low activity of the enzyme preparation.

^b Regression analysis was carried out of the nonsealed and sealed treatments over time and the significance of each regression coefficient (b) as well as the differences between the two regression coefficients. * is significant at 5% level.

was capable of simulating some of the sealing action, the firmness of soft fruit could be restored in aerated water, and that reduction of the RH inside the sealed enclosure cancelled these actions contradicts this argument. The indication is that these changes are not time-related senescence changes, but changes caused by the water stress.

Recently, water potential has been proposed to regulate active and passive transport by influencing the turgor pressure of cells (18, 35). Zimmermann's proposal (35) originates from the biphasic osmotic regulatory response of plants to salt and water stress. In the second phase of this response, the plant cells adjust their osmotic pressure in response to environmental stress. Thus, cells in higher plants have developed fascinating osmoregulatory mechanisms in order to withstand water stress. It is possible that the effects of sealing on leakage may relate to the second phase response of the fruit tissue to the higher turgor obtained by the restrained transpiration. The interrelationship between turgor and membrane may help explain the response to sealing in HDPE. In fact, Coster et al. (18) showed that significant changes in the thickness of cell membrane can occur as a result of (a) direct compression due to the turgor pressure, (b) indirect effect due to the stretching of cell walls, and (c) the stresses induced by the electric field in the membrane. Such changes in membrane thickness may provide the pressure-transducing mechanism required for osmoregulation. Such a model associated with changes occurring in the membrane could serve as the sensing mechanism of water stress. Indeed a change in membrane fluidity in response to osmotic concentration was demonstrated in liposomes (14). A good correlation was demonstrated in apple discs between the tonicity of the medium, ethylene production, and leakage of cellular content (17). However, with apples, leakage was promoted when turgor is increased in more than 5 bars by placing discs in hypotonic solutions. Placing discs in water or in hypotonic solutions delayed ripening in avocados (6).

Our work confirms previous reports showing that seal-packaging in plastic film delayed softening of all fruits tested (8). In fact, the extent of the inhibition of deterioration of the membrane integrity is not so marked as the inhibition of softening. Thus, 4-week-old sealed pepper fruit still had the firmness of freshly harvested fruit, but its membranes had already leaked amino acids at a rate higher than fresh fruit. However, the extent of delay of softening varies among different fruit. Thus, sealing of individual climacteric fruits in HDPE, such as tomato (12), avocado, mango, peach, and persimmon (Ben-Yehoshua et al., unpublished information), delayed softening but to only a small extent. The ripening process superseded and softening proceeded, even in sealed fruit, though at a slower rate. The control of the ripening process probably requires a more marked effect on CO₂, O₂, and ethylene than that individual sealing with thin HDPE film gives (21). However, with bell pepper, a nonclimacteric fruit, the sealed fruit did not soften; with lemon, another nonclimacteric fruit softening was very much inhibited. Softening in citrus fruit is of much smaller intensity than in climacteric fruit (27, 29, 30), but the breakdown of pectic materials in cell walls has been reported. Little has been published about pepper softening. Our work with lemons (unpublished information) also showed that free galacturonic acid and EDTA-soluble pectins rose gradually during storage. The amounts of insoluble pectins and cellulose did not change and the hemicellulose declined. Whereas neither of these changes occurred with sealed lemons during 4 months of storage at 14°C and 85% RH. Similar findings were also reported by Ben-Gad et al. (5).

The effects of sealing on polygalacturonase confirm previous reports of Van den Berg and Yang (34) that the activity of pectolytic enzymes is lower in WSA than in lower humidity regime of 90 to 95% RH.

The data on the similarity of CO₂ and ethylene production

and their endogenous concentration in lemons and in bell peppers support our early contention that sealing effects of these fruits could not be explained by the production of a new modified atmosphere around these fruits (8). These data verify our previous observation that CO₂ and O₂ concentrations in Shamouti oranges, Marsh grapefruit, and Eureka lemons were not affected by individual HDPE sealing (8, 10, 13). The rise in resistance to the gas diffusion was counteracted by an inhibition of the respiratory activity. However, for the present study with both lemons and bell peppers, this respiratory inhibition was not detected.

These results with lemons and peppers differ from other data demonstrating that, in many fruits and vegetables, sealing in plastic film delayed deterioration through modified atmosphere (21), that is, raising the CO₂ and lowering O₂ in their microatmosphere. In fact, our data with tomatoes showed that individual sealing in HDPE resulted in faster and more uniform development of red color which was due to the accumulated ethylene inside the sealed enclosure. It is tempting to make a conjecture that this water stress is more important than ethylene, O₂, and CO₂ with the lemon and pepper, both nonclimacteric fruits. Whereas with climacteric fruits, ethylene, O₂, and CO₂ play a major role.

Water stress accelerates the ripening process of avocados (1) and bananas (24). However, this finding could not always be repeated, even with the same fruit. Lizana (25) reported that the ripening of bananas was accelerated to a greater extent by WSA than by a RH of 85%. Thus, these data could not be properly summarized yet. Furthermore, the inactive role of ethylene in the mechanism of sealing action in delaying deterioration of lemon and pepper fruits is likely the exception rather than the rule which is that ethylene is greatly involved both as a cause and effect in several water stress phenomena. There are many reports of the enhancement of ethylene production by water stress (1, 9, 10, 22, 26, 28). Furthermore, ethylene-enhanced production was shown to be the cause of defoliation under water stress as an adaptation by the plant to resist water stress (9, 28). Generally speaking the involvement of the various plant hormones both in water stress and in membrane properties may indicate a possible role for them in the sensing mechanism for water stress (2, 10, 20, 22).

Acknowledgments—It is a pleasant duty to thank Irina Gero for her skillful and devoted technical assistance, V. Chew and J. Putter for their guidance in the statistical analyses, and S. Bullowa, E. Dorfman, and the secretarial staff of the United States Horticultural Research Laboratory for assistance in typing and editing.

LITERATURE CITED

- ADATO J, S GAZIT 1974 Water-deficit stress, ethylene production and ripening in avocado fruits. Plant Physiol 53: 45-46
- AHARONI N, S BEN-YEHOSHUA, AE RICHMOND 1975 Effects of water stress upon ethylene and endogenous content of abscisic acid and gibberellins in detached lettuce leaves (*Lactuca sativa* L.). Isr J Bot 24: 55
- BARRS HD, PE WEATHERBY 1962 A re-examination of the relative turgidity technique for estimation of water deficits in leaves. Aust J Biol Sci 15: 413– 428
- Ben-Arie R, S Lavee 1971 Pectin changes occurring in Elberta peaches suffering from wooly breakdown. Phytochemistry 10: 531-538
- BEN-GAD DY, EE GOLDSCHMIDT, SP MONSELISE 1981 Changes in cell wall components of 'Shamouti' (Citrus sinensis (L.) Osbeck) orange peel during maturation and senescence. Isr J Bot 30: 48
- BEN-YEHOSHUA S 1964 Respiration and ripening of the avocado fruit. Physiol Plant 77: 71-89
- BEN-YEHOSHUA S 1969 Gas exchange, transpiration and the commercial deterioration in storage of orange fruit. J Am Soc Hortic Sci 94: 524-528
- BEN-YEHOSHUA S 1978 Delaying deterioration of individual citrus fruits by seal packaging in film of high density polyethylene.
 General effects. Proc Int Soc Citriculture, Sydney, Australia, pp 110-115
- BEN-YEHOSHUA S, B ALONI 1974 Effects of water stress upon ethylene production by detached leaves of Valencia oranges. Plant Physiol 53: 863–865
- BEN-YEHOSHUA S, I KOBILER, B SHAPIRO 1979 Some physiological effects of delaying deterioration of citrus fruit by individual seal-packaging in highdensity polyethylene. J Am Soc Hortic Sci 104: 868-872

- BEN-YEHOSHUA S, I. KOBILER, B. SHAPIRO 1981 Effects of cooling versus sealpackaging with high density polyethylene on keeping qualities of various citrus cultivars. J Am Soc Hortic Sci 106: 536-540
- BEN-YEHOSHUA S, I KOBILER, B SHAPIRO 1979 Effect of seal packaging of individual tomatoes on rate of fruit deterioration. Prelim Rep Agric Res Orgn Bet Dagan 779
- BEN-YEHOSHUA S, B SHAPIRO, I KOBILER 1982 New method of degreening lemons by a combined treatment of ethylene-releasing agents and seal packaging in high-density polyethylene film. J Am Soc Hortic Sci 107: 365– 368
- BOROCHOV A, H BOROCHOV 1979 Increase in membrane fluidity in liposomes and plant protoplasts upon osmotic swelling. Biochim Biophys Acta 550: 546-549
- BOYER JS 1976 Water deficits and photosynthesis. In TT Kozlowski, ed, Water Deficits and Plant Growth, Vol. 4. Academic Press, New York, pp 153-190
- BRISTOW KL, WH VAN ZYL, JM DE KAGER 1981 Measurement of leaf water potential using the J14 press. J Exp Bot 32: 851-854
- BURG SP, KV THIMANN 1960 Studies of the ethylene production of apple tissue. Plant Physiol 35: 24-35
- 18. COSTER HGL, E STEUDLE, U ZIMMERMANN 1977 Turgor pressure sensing in plant cell membranes. Plant Physiol 58: 636-643
- DUBOIS M, KA GILLES, JK HAMILTON, PP REBERS, F SMITH 1956 Colorimetric method for determination of sugars and related substances. Anal Chem 28: 350-356
- GRIERSON W, WF WARDOWSKI 1978 Relative humidity effects on the postharvest life of fruits and vegetables. HortScience 13: 22-28
- HARDENBURG RF 1971 Effect of in-package environment on keeping quality of fruits and vegetables. HortScience 6: 190-201
- ITAI C, A BENZIONI 1976 B. Water stress and hormonal response. In OL Lange, L Kappen, ED Schulze, eds, Water and Plant Life, Ecological Studies, Vol

- 19. Springer-Verlag, Berlin, pp 225-242
- LEOPOLD A, M MUSGRAVE, K WILLIAMS 1981 Solute leakage resulting from leaf desiccation. Plant Physiol 68: 1222-1225
- LITTMANN MD 1972 Effect of water loss on the ripening of climacteric fruits.
 Queensl J Agric Anim Sci 29: 103-113
- LIZANA LA 1975 The influence of water stress and elevated temperatures on banana ripening. Proc Trop Reg Am Soc Hortic Sci 19: 137-145
- MAYAK S, AH HALEVY 1980 Flower senescence. In KV Thimann, ed, Plant Senescence. CRC Press, Boca Raton, FL, pp 131-156
- McCready R, E McComb 1952 Extraction and determination of total pectic materials in fruits. Anal Chem 24: 1986–1988
- MORGAN PW, WR JORDAN, TL DAVENPORT, JI DURHAM 1977 Abscission responses to moisture stress, auxin transport inhibitors and ethephon. Plant Physiol 59: 710-712
- 29. Riov K 1975 Polygalacturonase activity in citrus fruit. J Food Sci 40: 201-202
- ROUSE A, C ATKINS, E MOORE 1964 Seasonal changes occurring in the pectinesterase activity and pectin constituents of the component parts of citrus fruits. J Food Sci 29: 34-39
- SACHER J 1962 Relations between changes in membrane permeability and the climacteric in banana and avocado. Nature 195: 577-578
- SIMINOVITCH D, H THORRION, F GFELLER, H RHEAUME 1964 The quantitative estimation of frost injury and resistance in black locust, alfalfa and wheat tissues by determination of amino acids and other ninhydrin-reacting substances released after thawing. Can J Bot 42: 637-649
- UPDEGRAFF DM 1969 Semimicro determination of cellulose in biological materials. Anal Biochem 32: 420–424
- VAN DEN BERG L, SM YANG 1969 Effect of relative humidity on the production of extracellular pectolytic enzymes by Botrytis cinerea and Sclerotinia sclerotiorum. Can J Bot 47: 1007-1010
- 35. ZIMMERMANN U 1978 Physics of turgor- and osmoregulation. Annu Rev Plant Physiol 29: 121-148